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SZABO-SCANDIC HandelsgmbH

Quellenstraße 110, A-1100 Wien

T. +43(0)1 489 3961-0

F. +43(0)1 489 3961-7

mail@szabo-scandic.com

www.szabo-scandic.com

[linkedin.com/company/szaboscandic](https://www.linkedin.com/company/szaboscandic) 



Fc ϵ RI β shRNA (h) Lentiviral Particles: sc-45264-V

BACKGROUND

IgE Fc Receptor I binds to the Fc region of immunoglobulins ϵ chain with high affinity, and is responsible for initiating the allergic response. Binding of allergen to receptor-bound IgE leads to cell activation and the release of mediators such as histamines, responsible for the manifestations of allergy. IgE Fc Receptor I also induces the secretion of important lymphokines, effectors of the hypersensitivity response. It is a tetramer of a heavily glycosylated α chain, a β chain, and two disulfide linked γ chains. Structurally, the β chain contains four transmembrane regions with long cytoplasmic domains potentially involved in intracellular signaling. The cytoplasmic domains of the β and γ subunits each contain a conserved consensus sequence, ITAM, (immunoreceptor tyrosine activation motif). Phosphorylation of a pair of conserved tyrosine residues within this motif is required for signal transduction in mast cells and other hemopoietic cell types. A variant identified at Glu-237 of the β subunit has been implicated as a risk factor for atopic dermatitis and asthma.

REFERENCES

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- Shimizu, A. et al. 1988. Human and rat mast cell high-affinity immunoglobulin E receptors: characterization of putative α -chain gene products. *Proc. Natl. Acad. Sci. USA* 85: 1907-1911.
- Le Coniat, M. et al. 1990. The human genes for the α and γ subunits of the mast cell receptor for immunoglobulin E are located on human chromosome band 1q23. *Immunogenetics* 32: 183-186.
- Kuster, H. et al. 1992. The gene and cDNA for the human high affinity immunoglobulin E receptor β chain and expression of the complete human receptor. *J. Biol. Chem.* 267: 12782-12787.
- Maekawa, K. et al. 1992. Determination of the sequence coding for the β subunit of the human high-affinity IgE receptor. *FEBS Lett.* 302: 161-165.
- Penhallow, RC. et al. 1995. Temporal activation of nontransmembrane protein-tyrosine kinases following mast cell Fc ϵ RI engagement. *J. Biol. Chem.* 270: 23362-23365.

CHROMOSOMAL LOCATION

Genetic locus: MS4A2 (human) mapping to 11q12.1.

PRODUCT

Fc ϵ RI β shRNA (h) Lentiviral Particles is a pool of concentrated, transduction-ready viral particles containing 3 target-specific constructs that encode 19-25 nt (plus hairpin) shRNA designed to knock down gene expression. Each vial contains 200 μ l frozen stock containing 1.0×10^6 infectious units of virus (IFU) in Dulbecco's Modified Eagle's Medium with 25 mM HEPES pH 7.3. Suitable for 10-20 transductions. Also see Fc ϵ RI β siRNA (h): sc-45264 and Fc ϵ RI β shRNA Plasmid (h): sc-45264-SH as alternate gene silencing products.

STORAGE

Store lentiviral particles at -80° C. Stable for at least one year from the date of shipment. Once thawed, particles can be stored at 4° C for up to one week. Avoid repeated freeze thaw cycles.

APPLICATIONS

Fc ϵ RI β shRNA (h) Lentiviral Particles is recommended for the inhibition of Fc ϵ RI β expression in human cells.

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 μ l frozen viral stock containing 1.0×10^6 infectious units of virus (IFU); contains an shRNA construct encoding a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA.

GENE EXPRESSION MONITORING

Fc ϵ RI β (S-17): sc-33489 is recommended as a control antibody for monitoring of Fc ϵ RI β gene expression knockdown by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000) or immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500).

To ensure optimal results, the following support (secondary) reagents are recommended: 1) Western Blotting: use donkey anti-goat IgG-HRP: sc-2020 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible donkey anti-goat IgG-HRP: sc-2033 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use donkey anti-goat IgG-FITC: sc-2024 (dilution range: 1:100-1:400) or donkey anti-goat IgG-TR: sc-2783 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Fc ϵ RI β gene expression knockdown using RT-PCR Primer: Fc ϵ RI β (h)-PR: sc-45264-PR (20 μ l). Annealing temperature for the primers should be $55-60^\circ$ C and the extension temperature should be $68-72^\circ$ C.

BIOSAFETY

Lentiviral particles can be employed in standard Biosafety Level 2 tissue culture facilities (and should be treated with the same level of caution as with any other potentially infectious reagent). Lentiviral particles are replication-incompetent and are designed to self-inactivate after transduction and integration of shRNA constructs into genomic DNA of target cells.

RESEARCH USE

The purchase of this product conveys to the buyer the nontransferable right to use the purchased amount of the product and all replicates and derivatives for research purposes conducted by the buyer in his laboratory only (whether the buyer is an academic or for-profit entity). The buyer cannot sell or otherwise transfer (a) this product (b) its components or (c) materials made using this product or its components to a third party, or otherwise use this product or its components or materials made using this product or its components for Commercial Purposes.

PROTOCOLS

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